## Comparison of α-pinene and myrcene on attraction of mountain pine beetle, *Dendroctonus ponderosae* (Coleoptera: Scolytidae) to pheromones in stands of western white pine

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#### ABSTRACT

Multiple-funnel traps baited with *exo*-brevicomin and a mixture of *cis*- and *trans*-verbenol were used to test the relative attractiveness of myrcene and (-)- $\alpha$ -pinene to the mountain pine beetle, *Dendroctonus ponderosae* Hopkins, in a stand of western white pine, *Pinus monticola* Dougl. Traps baited with myrcene caught significantly more *D. ponderosae* than traps baited with (-)- $\alpha$ -pinene, irrespective of the presence of *exo*-brevicomin. *exo*-Brevicomin was attractive to *Thanasimus undatulus* (Say) (Coleoptera: Cleridae) whereas *Trypodendron lineatum* (Olivier) (Coleoptera: Scolytidae) was attracted to (-)- $\alpha$ -pinene. Our results support the use of myrcene in commercial trap lures and tree baits for *D. ponderosae* in stands of western white pine in British Columbia.

Key words: Scolytidae, Dendroctonus ponderosae, kairomones, Pinus monticola, Trypodendron lineatum, Cleridae, Thanasimus undatulus

#### **INTRODUCTION**

The mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Scolytidae), has killed over 500 million lodgepole, *Pinus contorta* var. *latifolia* Engelm., ponderosa, *P. ponderosa* P. Laws. and western white pines, *P. monticola* Dougl. (Pinaceae) in British Columbia over the past 80 years (Unger 1993). The current integrated pest management program for *D. ponderosae* in BC (Maclauchlan and Brooks 1994) is cost-effective, with positive economic, social and environmental impacts (Miller *et al.* 1993).

Semiochemicals play an important role in several tactics within the program (Maclauchlan and Brooks 1994). Population levels and flight periods of *D. ponderosae* are monitored with multiple-funnel traps baited with commercial lures consisting of the pheromones, *exo*-brevicomin and *cis*- and *trans*-verbenol, and the kairomone, myrcene (Stock 1984; Maclauchlan and Brooks 1994). The spread of infestations has been curtailed by the application of commercial tree baits consisting of the same semiochemicals (Borden and Lacey 1985; Borden *et al.* 1986) or simply the pheromones, *exo*-brevicomin and *cis*- and *trans*-verbenol, *exo*-brevicomin and *cis*- and *trans*-verbenol).

Semiochemical blends for these commercial lures and baits were developed in stands of lodgepole and ponderosa pine rather than western white pine, and discrepancies exist concerning the most appropriate kairomone. The host compound  $\alpha$ -pinene was more effective than myrcene in enhancing attraction of *D. ponderosae* to *trans*-verbenol in stands of western white pine in Idaho (Pitman 1971). Myrcene was more effective than  $\alpha$ -pinene in increasing attraction of *D. ponderosae* to pheromones in stands of ponderosa

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pine (Billings *et al.* 1976) and lodgepole pine (Borden *et al.* 1983; Conn *et al.* 1983). In lodgepole pine stands, catches of *D. ponderosae* in pheromone-baited traps exhibited a dose-dependent increase to both myrcene and 3-carene whereas  $\alpha$ -pinene had no effect (Miller and Borden 2000).

Our objective was to verify the effectiveness of myrcene, relative to  $\alpha$ -pinene, in commercial lures for *D. ponderosae* in stands of western white pine. Specifically we attempted to compare the response of beetles to pheromones in traps baited with  $\alpha$ -pinene to those baited with myrcene. Our expectation was that myrcene would be as effective as  $\alpha$ -pinene in attracting *D. ponderosae*.

#### **MATERIALS AND METHODS**

Semiochemical-Releasing Devices. Phero Tech Inc. (Delta, British Columbia) supplied polyethylene bubble-cap lures containing a 13:87 mixture of *trans*- and *cis*-verbenol [both chemical purities 98%; both enantiomeric compositions 83:17 (-):(+)], ( $\pm$ )-*exo*-brevicomin polyurethane flex lures (chemical purity >98%), and separate closed, low-density polyethylene bottles (15 mL) containing either  $\alpha$ -pinene [chemical purity >98%; enantiomeric composition > 99% (-)] or  $\beta$ -myrcene (chemical purity > 98%). The verbenols were released at a combined rate of approximately 1.74 mg/d at 24 °C (determined by weight loss) whereas  $\alpha$ -pinene and myrcene were released at approximately 413 mg/d and 281 mg/d at 24-28 °, respectively (determined by weight loss). *exo*-Brevicomin was released at approximately 0.1 mg/d at 24 °C (determined by collection of volatiles) (Phero Tech Inc.).

**Experiments.** Two experiments were conducted in a mature stand of western white pine with approximately 15% of live trees infested by *D. ponderosae* near Barriere, British Columbia (51°10'N, 120°8'W). In both experiments, forty 8-unit multiple-funnel traps (Lindgren 1983) (Phero Tech Inc.) were set 10-15 m apart, and  $\geq 2m$  from any tree, along two parallel transect lines spaced approximately 20 m apart. Each trap was suspended between trees by rope such that the top funnel of each trap was 1.3–1.5 m above ground. In Experiment 1, the effect of  $\alpha$ -pinene and myrcene on the attraction of *D. ponderosae* to traps baited with the verbenol mix was determined, with and without *exo*-brevicomin. All traps, baited with the verbenol mix, were set on 1 August 1990. The following treatments

were randomly assigned to 10 traps each: (1)  $\alpha$ -pinene; (2) myrcene; (3)  $\alpha$ -pinene and *exo*-brevicomin; and (4) myrcene and *exo*-brevicomin. Experiment 1 was terminated on 25 August 1990.

Experiment 2 tested the interaction between  $\alpha$ -pinene and myrcene on the attraction of *D. ponderosae* to traps baited with *exo*-brevicomin and the verbenol mix. All traps, baited with the verbenol mix and *exo*-brevicomin, were set on 25 August 1990. The following treatments were randomly assigned to 10 traps each: (1) no kairomone control; (2)  $\alpha$ -pinene; (3) myrcene; and (4)  $\alpha$ -pinene and myrcene. Experiment 2 was terminated on 12 September 1990.

Catches of *D. ponderosae* and serendipitous catches of *Trypodendron lineatum* (Olivier) (Coleoptera:Scolytidae) and *Thanasimus undatulus* (Say) (Coleoptera: Cleridae) were tallied for each treatment. Sexes of *D. ponderosae* captured in Experiment 2 were determined by dissection and examination of genitalia. Voucher specimens were deposited at the Entomology Museum, Simon Fraser University, Burnaby, BC.

Statistical Analyses. Trap catch data were analysed by 2-way ANOVA using the SYSTAT statistical package version 8.0 (SPSS 1998). The model factors in Experiment 1 were *exo*-brevicomin, monoterpenes ( $\alpha$ -pinene or myrcene), and the interaction between *exo*-brevicomin and monoterpenes. In Experiment 2, the model factors were  $\alpha$ -pinene,

myrcene and the interaction of  $\alpha$ -pinene and myrcene. Catches of *D. ponderosae* were transformed by ln(Y) to remove heteroscedasticity whereas catches of *Thanasimus* undatulus and *Trypodendron lineatum* were transformed by ln(Y+1) due to zero catches in some treatments. Sex ratio data, expressed as percentage of males in catches, from Experiment 2 were transformed by arcsine(Y). Fisher's least significant difference (LSD) multiple range tests were performed when P < 0.05.

#### **RESULTS AND DISCUSSION**

Our results clearly support the retention of myrcene in commercial lures for *D.* ponderosae in stands of western white pine. Catches of *D. ponderosae* were significantly higher in traps baited with myrcene than in traps baited with  $\alpha$ -pinene (Figs. 1,2). exo-Brevicomin did not affect the preference of beetles for myrcene over  $\alpha$ -pinene ( $F_{1,36} =$ 0.605, P = 0.442) in Experiment 1 nor was there any interaction between  $\alpha$ -pinene and myrcene on catches of *D. ponderosae* ( $F_{1,36} = 0.018$ , P = 0.894) in Experiment 2. There was no significant difference in sex ratio among the treatments in Experiment 2 ( $F_{3,19} =$ 2.232, P = 0.121) with the mean ( $\pm$  SE) percentage of males in catches at 55  $\pm$  3 %.



**Figure 1.** Effect of *exo*-brevicomin (eB),  $\alpha$ -pinene (P) and myrcene (M) on the attraction of *Dendroctonus ponderosae* to verbenol-baited multiple-funnel traps from 1 August to 25 August 1990 (n = 10). Means followed by different letters are significantly different at P < 0.05 (LSD test).



# Mean (+SE) number of beetles

**Figure 2.** Effect of  $\alpha$ -pinene (P) and myrcene (M) on the attraction of *Dendroctonus ponderosae* to multiple-funnel traps baited with verbenols and *exo*-brevicomin from 25 August to 12 September 1990 (n = 10). Means followed by different letters are significantly different at P < 0.05 (LSD test); control (c).

Our results are inconsistent with those of Pitman (1971) who demonstrated that  $\alpha$ pinene was more effective than myrcene in attracting *D. ponderosae* in stands of western white pine and are surprising since  $\alpha$ -pinene is the most common monoterpene in the resin of western white pine, which has low amounts of myrcene (Mirov 1961). The relative proportion of myrcene is higher in the resin of lodgepole and ponderosa pines with amounts of myrcene greater than or equal to amounts of  $\alpha$ -pinene (Mirov 1961; Shrimpton 1973). Geographic variation in semiochemical responses, similar to that in *Ips pini* (Say) (Miller *et al.* 1997), may explain some of these results.

Finally, research by Pitman (1971), Billings *et al.* (1976), Borden *et al.* (1983) and Conn *et al.* (1983) were conducted before the importance of the enantiomeric composition of  $\alpha$ -pinene was widely recognised. It is likely, but not certain, that they used either (±)- or (-)- $\alpha$ -pinene due to the high costs associated with (+)- $\alpha$ -pinene. We used (-)- $\alpha$ -pinene in our trials since it is the predominant enantiomer in the resin of western white pine phloem tissue (Mirov 1961).

In Experiment 1, catches of *Trypodendron lineatum* were lowest in traps baited with myrcene alone, and highest in traps baited with either  $\alpha$ -pinene or *exo*-brevicomin (Table 1).  $\alpha$ -Pinene significantly increases the attraction of *T. lineatum* to ethanol and the

pheromone lineatin (Borden *et al.* 1982; Schroeder and Lindelöw 1989). No *T. lineatum* were caught in Experiment 2.

Table 1

Mean $(\pm SE)$ catches of <i>Trypoder</i>	<i>ndron lineatum</i> (Scolytidae	e) and Thanasimus undatulus
(Cleridae) in verbenol-baited multiple-funnel traps from 1 August to 25 August 1990 <sup>a</sup> .		
Treatment	Trypodendron lineatum	Thanasimus undatulus
(-)-α-Pinene	$11 \pm 5 b$	$1 \pm 1$ a
Myrcene	$3 \pm 1 a$	$1 \pm 1 a$
$(-)-\alpha$ -Pinene + $(\pm)$ - <i>exo</i> -brevicomin	$12 \pm 2 b$	$28 \pm 5 c$
Myrcene + $(\pm)$ - <i>exo</i> -brevicomin	9 ± 2 b	16 ± 3 b

<sup>*a*</sup> Means within the same column followed by the same letter are not significantly different, P < 0.05 (LSD multiple comparison test).

The predator, *Thanasimus undatulus*, showed a preference for traps baited with *exo*brevicomin in combination with myrcene or  $\alpha$ -pinene, particularly the latter (Table 1). As might be expected for a generalist predator, similar results with *T. undatulus* have been reported with the following bark beetle pheromones: frontalin, *exo-* and *endo-*brevicomin, ipsdienol, ipsenol, and *cis-*verbenol (Kline *et al.* 1974; Dyer 1975; Chatelain and Schenk 1984; Miller *et al.* 1987; Miller and Borden 1990; Miller *et al.* 1991; Miller *et al.* 1997; Poland and Borden 1997). Usually, *T. undatulus* are not attracted to host tree compounds (Furniss and Schmitz 1971; Miller and Borden 1990) although Macías-Sámano *et al.* (1998) demonstrated attraction of *T. undatulus* to host blends from grand fir, *Abies grandis* (Dougl.) Lindl. No *T. undatulus* were caught in Experiment 2.

#### ACKNOWLEDGEMENTS

We thank J.H. Borden, K.O. Britton, B. Sullivan and an anonymous reviewer for their reviews of the manuscript. L.E. Maclauchlan, J.E. Macías-Sámano and D. Piggin provided technical and field assistance. This research was supported by the Science Council of British Columbia.

### REFERENCES

- Billings, R.F., R.I. Gara and B.F. Hrutfiord. 1976. Influence of ponderosa pine resin volatiles on the response of *Dendroctonus ponderosae* to synthetic *trans*-verbenol. Environmental Entomology 5: 171-179.
- Borden, J.H. and T.E. Lacey. 1985. Semiochemical-based manipulation of the mountain pine beetle, *Dendroctonus ponderosae* Hopkins: a component of lodgepole pine silviculture in the Merritt Timber Supply Area of British Columbia. Zeitschrift für angewandte Entomologie 99: 139-145.
- Borden, J.H., C.J. King, S. Lindgren, L. Chong, D.R. Gray, A.C. Oehlschlager, K.N. Slessor and H.D. Pierce, Jr. 1982. Variation in response of *Trypodendron lineatum* from two continents to semiochemicals and trap form. Environmental Entomology 11: 403-408.
- Borden, J.H., J.E. Conn, L.M. Friskie, B.E. Scott and L.J. Chong. 1983. Semiochemicals for the mountain pine beetle, *Dendroctonus ponderosae* (Coleoptera: Scolytidae), in British Columbia: baited-tree studies. Canadian Journal of Forest Research 13: 325-333.
- Borden, J.H., L.J. Chong and T.E. Lacey. 1986. Pre-logging baiting with semiochemicals for the mountain pine beetle, *Dendroctonus ponderosae*, in high hazard stands of lodgepole pine. Forestry Chronicles 62: 20-23.
- Borden, J.H., L.J. Chong, B.S. Lindgren, E.J. Begin, T.M. Ebata, L.E. Maclauchlan and R.S. Hodgkinson. 1993. A simplified tree bait for the mountain pine beetle. Canadian Journal of Forest Research 23: 1108-1113.

- Chatelain, M.P. and J.A. Schenk. 1984. Evaluation of frontalin and exo-brevicomin as kairomones to control mountain pine beetle (Coleoptera: Scolytidae) in lodgepole pine. Environmental Entomology 13: 1666-1674.
- Conn, J.E., J.H. Borden, B.E. Scott, L.M. Friskie, H.D. Pierce, Jr. and A.C. Oehlschlager. 1983. Semiochemicals for the mountain pine beetle, *Dendroctonus ponderosae* (Coleoptera: Scolytidae) in British Columbia: field trapping studies. Canadian Journal of Forest Research 13: 320-324.
- Dyer, E.D.A. 1975. Frontalin attractant in stands infested by the spruce beetle, *Dendroctonus rufipennis* (Coleoptera: Scolytidae). The Canadian Entomologist 107: 979-988.
- Furniss, M.M. and R.F. Schmitz. 1971. Comparative attraction of Douglas-fir beetles to frontalin and tree volatiles. U.S. Department of Agriculture Forest Service Research Paper INT-96.
- Kline, L.N., R.F. Schmitz, J.A. Rudinsky and M.M. Furniss. 1974. Repression of spruce beetle (Coleoptera ) attraction by methylcyclohexenone in Idaho. The Canadian Entomologist 106: 485-491.
- Lindgren, B.S. 1983. A multiple-funnel trap for scolytid beetles. The Canadian Entomologist 115: 299-302.
- Macías-Sámano, J.E., J.H. Borden, R. Gries, H.D. Pierce, Jr., G. Gries and G.G.S. King. 1998. Primary attraction of the fir engraver, *Scolytus ventralis*. Journal of Chemical Ecology 24: 1049-1075.
- Maclauchlan, L.E. and J.E. Brooks. 1994. Strategies and tactics for managing the mountain pine beetle, *Dendroctonus ponderosae*. Kamloops Forest Region, British Columbia Forest Service. 60 pp.
- Miller, D.R. and J.H. Borden. 1990. β-Phellandrene: kairomone for pine engraver, *Ips pini* (Say) (Coleoptera: Scolytidae). Journal of Chemical Ecology 16: 2519-2531.
- Miller, D.R. and J.H. Borden. 2000. Dose-dependent and species-specific responses of pine bark beetles (Coleoptera: Scolytidae) to monoterpenes in association with pheromones. The Canadian Entomologist 132: 183-195.
- Miller, D.R., J.H. Borden, G.G.S. King and K.N. Slessor. 1991. Ipsenol: an aggregation pheromone for *Ips latidens* (LeConte) (Coleoptera: Scolytidae). Journal of Chemical Ecology 17: 1517-1527.
- Miller, D.R., J.A. Carlson and M. Stemeroff. 1993. Socioeconomic analysis of mountain pine beetle management in British Columbia. British Columbia Forest Service. 67 pp.
- Miller, D.R., K.E. Gibson, K.F. Raffa, S.J. Seybold, S.A. Teale and D.L. Wood. 1997. Geographic variation in response of pine engraver, *Ips pini*, and associated species to pheromone, lanierone. Journal of Chemical Ecology 23: 2013-2031.
- Miller, M.C., J.C. Moser, M. McGregor, J.C. Gregoire, M. Baisier, D.L. Dahlsten and R.A. Werner. 1987. Potential for biological control of native North American *Dendroctonus* beetles (Coleoptera: Scolytidae). Annals of the Entomological Society of America 80: 417-428.
- Mirov, N.T. 1961. Composition of gum turpentines of pines. U.S. Department of Agriculture Forest Service Technical Bulletin No. 1239.
- Pitman, G.B. 1971. *trans*-Verbenol and alpha-pinene: their utility in manipulation of the mountain pine beetle. Journal of Economic Entomology 64: 426-430.
- Poland, T.M. and J.H. Borden. 1997. Attraction of a bark beetle predator, *Thanasimus undatulus* (Coleoptera: Cleridae), to pheromones of the spruce beetle and two secondary bark beetles (Coleoptera: Scolytidae). Journal of the Entomological Society of British Columbia 94: 35-41.
- Schroeder, L.M. and Å. Lindelöw. 1989. Attraction of scolytids and associated beetles by different absolute amounts and proportions of α-pinene and ethanol. Journal of Chemical Ecology 15: 807-817.
- Shrimpton, D.M. 1973. Extractives associated with wound response of lodgepole pine attacked by the mountain pine beetle and associated microorganisms. Canadian Journal of Botany 51: 527-534.
- Stock, A.J. 1984. Use of pheromone baited Lindgren funnel traps for monitoring mountain pine beetle flights. British Columbia Forest Service. 11 pp.
- SPSS Inc. 1998. SYSTAT 8.0 Statistics. Chicago, IL. 1086 pp.
- Unger, L. 1993. Mountain pine beetle. Forestry Canada Forest Pest Leaflet 76.